

NISTTech

Micropatterning of Extracellular Matrix Proteins Using Micro Photo Ablation of Poly Vinyl Alcohol (PVA) Monolayers

Creates patterns of extracellular matrix proteins in live cell applications

Description

This process creates submicron patterns of extracellular matrix proteins (EMP) in a cellular environment to permit the study of how live cells interact with these proteins of interest. Tunable multi-photon lasers photo ablate (remove thin layers) of poly vinyl alcohol (PVA) leaving behind tiny micropatterns. 'Region of interest' (ROI) software functions precisely control the size and shapes of the patterns on which EMPs will be deposited. Many different proteins can be built up in the same region to provide more information on cellular interactions.

Applications

- **Microbiology**
Study extracellular matrix interactions among cells and their effects on cellular functioning and regulation

Advantages

- **'Region of interest' functions expand speed and flexibility**
The region of interest functions create easily modified "virtual masks" in any shape or pattern. Creates up to several hundred patterns within 30 seconds
- **Micro photo ablation provides more 'live cell' abilities**
Allows live cell imaging of multiple fluorophores. Works with total internal reflection fluorescence (TIRF) microscopy Allows kinetic quantification of ECM-cell interactions

Abstract

The advent of micro-lithography techniques into the study of cell biology have greatly increased our understanding of how cellular functioning is in part regulated by the interaction of cells with the extracellular matrix (ECM). Currently many techniques have used micro-contact patterning (MCP) to apply ECM proteins in distinct localized patterns. These techniques require the fabrication of silicone-based stamps to either "ink" proteins directly or indirectly onto a gold coated surface, limiting the user to a specified stamp shape and size.

To bypass the necessity of a physical stamp we have devised a technique to generate submicron sized spots using a tunable multi-photon laser coupled to a confocal microscope to photo ablate hydrophilic poly vinyl alcohol (PVA) hydro gels and self assembled monolayers. Through controlled photo ablation, PVA layers are locally removed allowing deposition of ECM proteins into distinct patterns. By using a software-controlled region of interest (ROI) function, we are able to precisely control patterns, generating up to several hundred within 30 seconds. The use of ROI's produces a "virtual mask" that can be created in any shape or pattern and is easily modified. Unlike MCP techniques, micro photo ablation (MPA) allows live cell imaging of multiple

fluorophores and is possible even with total internal reflection fluorescence (TIRF) microscopy. In addition, MPA allows kinetic quantification of ECM-cell interactions. This technique uses a self-assembled monolayer (SAMs) model for application together with localized photo ablation allows the versatility to create protein spots of any size or shape easily on the same cover slip. Furthermore, this process can be repeated multiple times to directly conjugate different proteins to the same local region allowing the investigation of how single cells probe their surroundings to discern different ECM proteins.

Inventors

- Wang, Francis W.
- Doyle, Andrew D.
- Yamada, Kenneth M.

Citations

1. A.D. Doyle, F.W. Wang, K. Matsumoto, K.M. Yamada, One dimensional topography underlies three dimensional cell migration, Journal of Cell Biology vol. 184 no. 4 481-490 (2009)

References

- U.S. Patent Application 20090096133
- Docket: 08-036

Status of Availability

This invention is available for licensing.

Last Modified: 02/11/2011